

## ANTIOXIDANT PROPERTIES OF DIFFERENT APPLE CULTIVARS

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## ABSTRACT

Dietary antioxidants can be an important part of the healthy diet. Polyphenols from the commonly consumed apple can be possible sources of intake of these phytochemicals.

In our study, the antioxidant properties of six apple cultivars (Golden Delicious, Granny Smiths, Idared, Jonagold, Jonagored and Mutsu) harvested at commercial maturity were examined. Flesh and skin were separated and total phenolics by the Folin-Ciocalteu assay, ferric reducing antioxidant power (FRAP) and radical scavenging activities using DPPH method were determined for each. For all apples, polyphenol content and antioxidant activity of the skin were significantly higher than that of the flesh showing that apple peel is a valuable part of the fruit. There was a good linear correlation between the polyphenol content and FRAP showing that mainly polyphenols are responsible for this type of antioxidant reaction. Regarding the cultivars, there was an obvious difference between the antioxidant activities of the examined apples. The green variety Granny Smith showed the best results followed by the red-skin apples while yellow-skin apples had the lowest activity.

**Keywords:** antioxidants, apple cultivars, polyphenols, FRAP

## INTRODUCTION

Fruits are the main sources of dietary antioxidants (KAUR AND KAPOOR, 2001). They are rich in polyphenols, vitamins, organic acids, and minerals. Apple (*Malus domestica*) is one of the most widely consumed fruit, has a long history of cultivation and more than 7000 cultivars. According to an American study, apple has the second highest antioxidant activity among the most frequently consumed fruits (BOYER AND LIU, 2014), only cranberry showed higher activity than apple. The amount and type of polyphenols differ among apple cultivars but the most frequently determined phenolics are quercetin glycosids, catechin and epicatechin, proanthocyanidins, chlorogenic, gallic, coumaric acids and floridzin (FRANCINI AND SEBASTIANI, 2013). In this study extracts from the flesh and peel of 6 apple cultivars were investigated for their polyphenol content and antioxidant activity.

## MATERIAL AND METHOD

## Apple cultivars

The apple cultivars used were: Golden Delicious, Granny Smiths, Idared, Jonagold, Jonagored, and Mutsu. The cultivars were purchased from the same apple orchard near Pécs, Hungary, and were harvested at their marketable maturity stage.

## Preparation of the samples

Apples were peeled and flesh and peel were separately lyophilized (Lyovac GT2, Germany), then grounded to fine powder.

### Extraction of polyphenols

One gram of lyophilized apple powder was extracted for 24 h in 20 ml 50 % ethanol at room temperature, then the samples were centrifuged (1000 g, 30 min) and the supernatants were used for further investigations.

### Determination of total polyphenols by the Folin-Ciocalteu assay

Extracts were diluted five times with 50% ethanol and 0.2 ml of diluted extracts were mixed with 0.2 ml ethanol (96 %), 1 ml distilled water and 0.1 ml of Folin-Ciocalteu's reagent. After 5 min, 0.2 ml sodium carbonate solution (5 %) was added to the mixtures, then incubated at room temperature in dark for 1 hour. The developed green colour was measured at 725 nm by an UV/VIS spectrophotometer (Philips PU8740). Polyphenol content was expressed in mg gallic acid equivalent (GAE)/1 g solid (lyophilized apple).

### Determination of radical scavenging activity with the DPPH assay

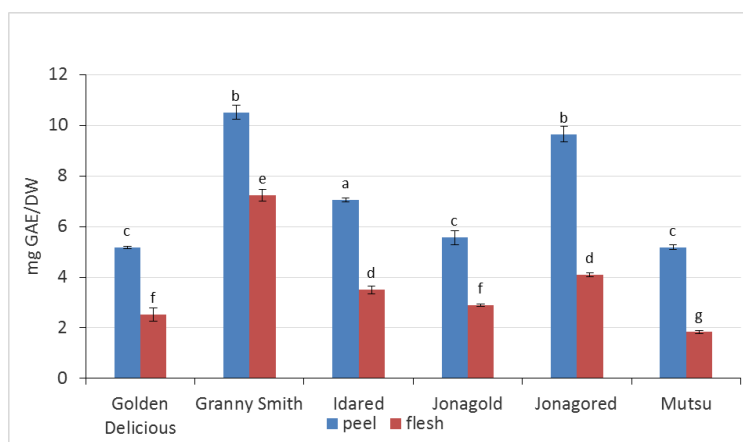
For this measurement, the same 5-fold dilutions as for polyphenol determination were used. 1.2 ml of 100  $\mu$ M DPPH (2,2-diphenyl-1-picrylhydrazil) ethanolic solution was added to 0.2 ml diluted sample and this mixture was incubated in the dark at room temperature for 30 min. In the control ethanol was added instead of the sample. After 30 min the changes in colour (from violet to yellow) were measured at 517 nm. Antioxidant activity was calculated by the following equation: DPPH• scavenging effect (%) =  $((A_c - A_s)/A_c) \times 100$ ,

where:  $A_c$  was the absorbance of the control and  $A_s$  the absorbance of the apple sample.

### Ferric reducing antioxidant power assay (FRAP)

FRAP solution contained 80 ml of 300 mM acetate buffer (pH 3.6); 8 ml of 10 mM 2,4,6-Tri(2-pyridyl)-s-triazine (TPTZ; Sigma-Aldrich, Germany) diluted in 40 mM hydrochloric acid; 8 ml of 20 mM iron(III) chloride and 4.8 ml of distilled water. In the reaction mixture 4 ml FRAP reagent was added to 120  $\mu$ l 5-fold diluted sample. This mixture was incubated at 37 °C for 30 min and then absorbance was measured at 593 nm. Calibration was made using 1 mM iron(II) sulfate solution in the concentration range of 0.1-1.0 mM. The FRAP of the extracts was expressed as  $\mu$ M Fe(II)/g lyophilized apple.

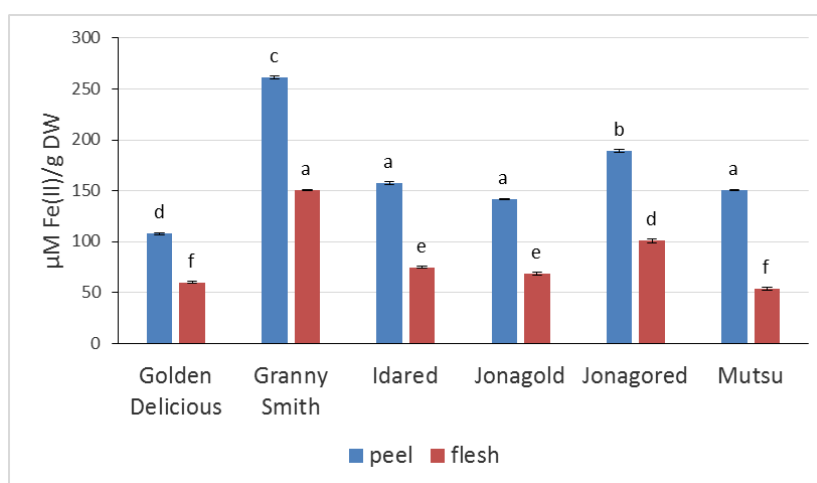
## RESULTS AND DISCUSSION



**Figure 1. Total polyphenol content in the peel and flesh of the investigated apple cultivars. Different small letters represent significant differences ( $p < 0.05$ )**

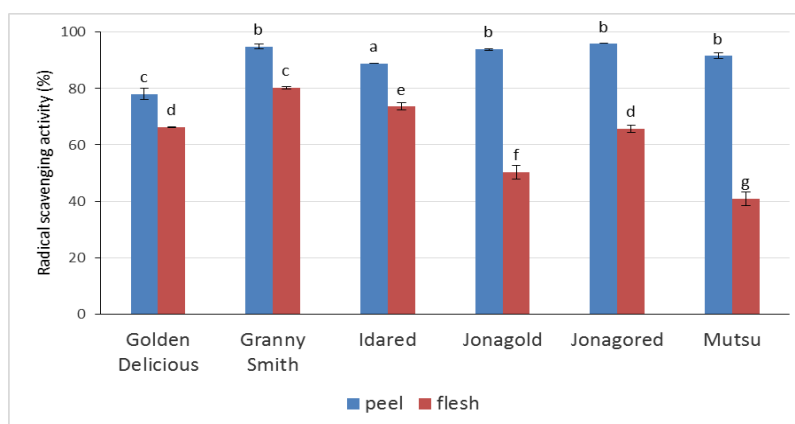
Total polyphenols have been found in the highest amount in the peel of the Granny Smith cultivar, followed by the peel of Jonagored (*Figure 1*).

In general polyphenolics in peels were represented in half to twice higher amount, than in the flesh. WOLFE AND CO-WORKERS (2003) found also that some apple peels contain from two to six times more phenolic compounds, and two to three times more flavonoids in the peels than in the flesh. Because of the skin is the first defense line in the fruits it is not surprising that polyphenols with antioxidant and antimicrobial activity are concentrated here. According to HUBER AND RUPASINGHE (2009) some compounds like catechin, procyanidin, epi-catechin, and phloridzin are in much lower concentrations in the flesh than in the peels. Quercetin conjugates were found exclusively in the peel of the apples (BOYER AND LIU, 2004). Similar to the polyphenol content antioxidant activity (FRAP) was also highest in the peel of Granny Smith cultivar, followed by the peel of Jonagored (*Figure 2*).



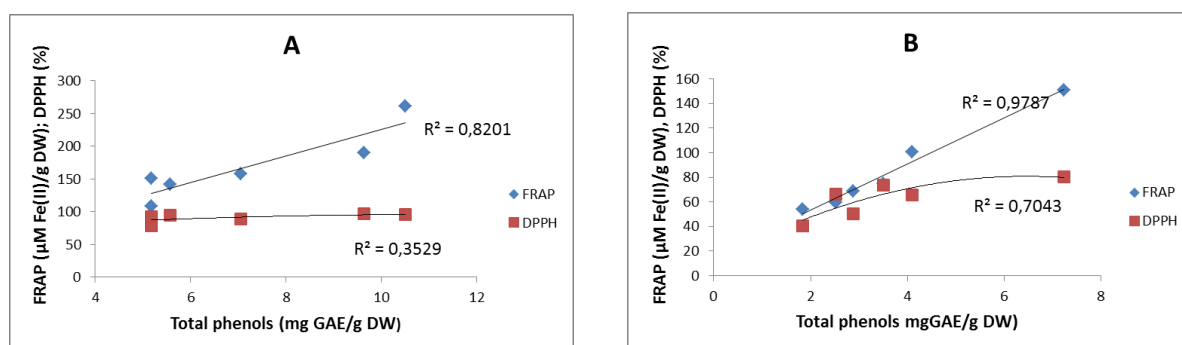
**Figure 2. Ferric reducing power (FRAP) in the peel and flesh of the investigated apple cultivars. Different small letters represent significant differences ( $p < 0.05$ )**

In spite of this the radical scavenging activity seems not to depend on the polyphenol content and showed also limited correlation with the type of cultivar, but strong correlation with from what part of the fruit, peel or flesh, was the sample taken (*Figure 3*). In this case the radical scavenging activity was much higher in the peel than in the flesh.



**Figure 3. Radical scavenging activity (%) in the peel and flesh of the investigated apple cultivars. Different small letters represent significant differences ( $p < 0.05$ )**

Ferric reducing antioxidant power was also higher in the peels compared to the flesh of the apples. The antioxidant activity was also much greater in the peels when compared to the flesh, depending on the variety of the apple. There was a good linear correlation between the polyphenol content and FRAP, but a weak, non-linear correlation between polyphenol content and radical scavenging activity in the case of the peels, and a stronger, non-linear correlation in the case of flesh (Figure 4).



**Figure 4. Correlation between total phenol content and ferric reducing power, and radical scavenging activity in the peel (A) and flesh (B) of the investigated apple cultivars.**

## CONCLUSIONS

The polyphenol content and antioxidant capacity differed significantly among the different apple cultivars and the green-coloured Granny Smiths proved to be the best. In all cases, peels had much higher amount of phenolics and greater antioxidant activity than the flesh, showing that apple peel is a valuable part of the fruit.

## ACKNOWLEDGEMENTS

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